UNITED STATES PATENT AND TRADEMARK OFFICE

UNITED STATES DEPARTMENT OF COMMERCE
United States Patent and Trademark Office
Address: COMMISSIONER FOR PATENTS
P.O. Box 1450
Alexandria, Virginia 22313-1450
www.uspto.gov

| APPLICATION NO. | FILING DATE | FIRST NAMED INVENTOR | ATTORNEY DOCKET NO. | CONFIRMATION NO. |
|---|---|---|---|---|
| 10/673,786 | 09/30/2003 | Valery Zavenovich Akhverdian | US-115 | 7880 |

38108 7590 01/18/2007

CERMAK & KENEALY LLP
ACS LLC
515 EAST BRADDOCK ROAD
SUITE B
ALEXANDRIA, VA 22314

| EXAMINER | |
|---|---|
| RAMIREZ, DELIA M | |
| **ART UNIT** | **PAPER NUMBER** |
| 1652 | |

| SHORTENED STATUTORY PERIOD OF RESPONSE | MAIL DATE | DELIVERY MODE |
|---|---|---|
| 3 MONTHS | 01/18/2007 | PAPER |

**Please find below and/or attached an Office communication concerning this application or proceeding.**

If NO period for reply is specified above, the maximum statutory period will apply and will expire 6 MONTHS from the mailing date of this communication.

| Office Action Summary | Application No. | Applicant(s) | |
|---|---|---|---|
| | 10/673,786 | AKHVERDIAN ET AL. | |
| | Examiner | Art Unit | |
| | Delia M. Ramirez | 1652 | |

*-- The MAILING DATE of this communication appears on the cover sheet with the correspondence address --*

**Period for Reply**

A SHORTENED STATUTORY PERIOD FOR REPLY IS SET TO EXPIRE 3 MONTH(S) OR THIRTY (30) DAYS, WHICHEVER IS LONGER, FROM THE MAILING DATE OF THIS COMMUNICATION.

- Extensions of time may be available under the provisions of 37 CFR 1.136(a). In no event, however, may a reply be timely filed after SIX (6) MONTHS from the mailing date of this communication.
- If NO period for reply is specified above, the maximum statutory period will apply and will expire SIX (6) MONTHS from the mailing date of this communication.
- Failure to reply within the set or extended period for reply will, by statute, cause the application to become ABANDONED (35 U.S.C. § 133). Any reply received by the Office later than three months after the mailing date of this communication, even if timely filed, may reduce any earned patent term adjustment. See 37 CFR 1.704(b).

**Status**

1)☒ Responsive to communication(s) filed on *19 October 2006*.

2a)☒ This action is **FINAL**. 2b)☐ This action is non-final.

3)☐ Since this application is in condition for allowance except for formal matters, prosecution as to the merits is closed in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11, 453 O.G. 213.

**Disposition of Claims**

4)☒ Claim(s) *12,15,16,19 and 21-24* is/are pending in the application.

4a) Of the above claim(s) _____ is/are withdrawn from consideration.

5)☐ Claim(s) _____ is/are allowed.

6)☒ Claim(s) *12,15,16,19 and 21-24* is/are rejected.

7)☐ Claim(s) _____ is/are objected to.

8)☐ Claim(s) _____ are subject to restriction and/or election requirement.

**Application Papers**

9)☐ The specification is objected to by the Examiner.

10)☐ The drawing(s) filed on _____ is/are: a)☐ accepted or b)☐ objected to by the Examiner.

Applicant may not request that any objection to the drawing(s) be held in abeyance. See 37 CFR 1.85(a).

Replacement drawing sheet(s) including the correction is required if the drawing(s) is objected to. See 37 CFR 1.121(d).

11)☐ The oath or declaration is objected to by the Examiner. Note the attached Office Action or form PTO-152.

**Priority under 35 U.S.C. § 119**

12)☒ Acknowledgment is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d) or (f).

a)☒ All b)☐ Some * c)☐ None of:

1.☒ Certified copies of the priority documents have been received.

2.☐ Certified copies of the priority documents have been received in Application No. _____.

3.☐ Copies of the certified copies of the priority documents have been received in this National Stage application from the International Bureau (PCT Rule 17.2(a)).

* See the attached detailed Office action for a list of the certified copies not received.

**Attachment(s)**

1) ☐ Notice of References Cited (PTO-892)
2) ☐ Notice of Draftsperson's Patent Drawing Review (PTO-948)
3) ☐ Information Disclosure Statement(s) (PTO/SB/08) Paper No(s)/Mail Date _____.
4) ☐ Interview Summary (PTO-413) Paper No(s)/Mail Date. _____.
5) ☐ Notice of Informal Patent Application
6) ☐ Other: _____.

## DETAILED ACTION

### *Status of the Application*

Claims 12, 15-16, 19, 21-24 are pending.

Applicant's amendment of claims 12, 15, 19, 21-22, addition of claims 23-24, and cancellation of claims 13-14, 17-18, 20 as submitted in a communication filed on 10/19/2006 is acknowledged.

Rejections and/or objections not reiterated from previous office actions are hereby withdrawn.

### *Claim Rejections - 35 USC § 112, First Paragraph*

1. The text of those sections of Title 35, U.S. Code not included in this action can be found in a prior Office action.

2. Claims 12, 19, 21-22 remain rejected under 35 U.S.C. 112, first paragraph, as failing to comply with the written description requirement. The claim(s) contains subject matter which was not described in the specification in such a way as to reasonably convey to one skilled in the relevant art that the inventor(s), at the time the application was filed, had possession of the claimed invention.

3. Applicant argues that claim 12 has been amended to specify the amino acid sequence of the aspartate aminotransferase of the method, limit the source of the thrABC and rhtA genes, and specify that the bacterium is modified by increasing the copy number of the gene or modifying an expression control sequence of the gene.

4. Applicant's arguments have been fully considered but are not deemed persuasive to overcome the instant rejection. Claims 12, 19, 21-22 as amended are directed to a method for the production of L-threonine wherein said method comprises cultivating an L-threonine-producing *Escherichia coli* strain which has been modified to increase the expression of a gene encoding the polypeptide of SEQ ID NO: 2, and wherein said increase in expression is achieved by any modification in an expression control sequence of said gene. The Examiner acknowledges the amendments to the claims, which overcome

some of the grounds of rejections previously applied. However, the claims still require any modification made to the regulatory region of the gene. While it is agreed that the use of strong heterologous promoters is well known and widely used in the art, the claims required unknown modifications in the gene which would result in increased expression, such as mutations in the endogenous promoter. There is no disclosure of any additional modifications in the regulatory region of the gene which would result in increased expression. There is no structure/function correlation which would allow one of skill in the art to recognize which structural modifications need to be made to achieve enhanced expression of the gene. Thus, one cannot reasonably conclude that the genus of modifications encompassed by the claims is adequately described by the teachings of the specification.

5. Claims 12, 19, 21-22 remain rejected under 35 U.S.C. 112, first paragraph, because the specification, while being enabling for a method for the production of L-threonine wherein said method comprises cultivating an L-threonine-producing *Escherichia coli* strain which has been modified to increase the expression of a polynucleotide encoding the polypeptide of SEQ ID NO: 2, and wherein said increase in expression is achieved by increasing the copy number of said polynucleotide or by using a heterologous promoter, does not reasonably provide enablement for a method for the production of L-threonine wherein said method comprises cultivating an L-threonine-producing *Escherichia coli* strain which has been modified to increase the expression of a gene encoding the polypeptide of SEQ ID NO: 2, and wherein said increase in expression is achieved by any modification in an expression control sequence of said gene. The specification does not enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to make and/or use the invention commensurate in scope with these claims.

6. Applicant argues that claim 12 has been modified to specify that the increase in expression is due to either an increase in the copy number, or by modification of an expression control sequence of the gene.

7. Applicant's arguments have been fully considered but are not deemed persuasive to overcome the instant rejection. The Examiner acknowledges the amendments made which now limit the genus of genes encoding the aspartate aminotransferase to those encoding the polypeptide of SEQ ID NO: 2, as well as the source of the thrABC and rhtA genes to those from *E. coli*. The Examiner also acknowledges that replacement of the endogenous promoter of a gene with a heterologous promoter which would increase expression of said gene is well known in the art. However, the claims as written encompass unknown modifications within the regulatory region of the gene which would result in increased expression. As previously indicated in the Non Final action mailed on 5/19/2006, these modifications as recited comprise, for example, mutations in the endogenous promoter of the gene. The specification is silent with regard to the structural modifications required in the regulatory region of a gene which would result in increased expression other than the use of strong heterologous promoters known in the art. The art does not provide any information as to which are the structural modifications in the regulatory region of a gene encoding the polypeptide of SEQ ID NO: 2 which would result in increased expression. In the absence of some knowledge or guidance as to which structural changes in the regulatory region of a gene would most likely result in increased expression, determining which modifications in the regulatory region of a gene will result in increased expression would require undue experimentation. Thus, one cannot reasonably conclude that the claimed invention is fully enabled by the teachings of the specification or the art.

***Claim Rejections - 35 USC § 103***

8. The text of those sections of Title 35, U.S. Code not included in this action can be found in a prior Office action.

9. Claims 12, 15-16, 19, 21-22 remain rejected and new claims 23-24 are rejected under 35 U.S.C. 103(a) as being unpatentable over Katsumata et al. (EP 0219027 published 4/22/1987; cited in the IDS) in view of Debabov et al. (U.S. Patent No. 5175107 issued on 12/29/1992; cited in the IDS), Edwards et al. (WO 87/00202 published on 1/15/1987; cited in the IDS), and further in view of Kishino et al. (U.S. Patent No. 6319696 issued on 11/20/2001). This rejection has been discussed at length in the Non Final action mailed on 5/19/2006. It is now applied to new claims 23-24 for the reasons of record and those set forth below.

10. Applicant traverses the rejection on the grounds that (1) there are many aminotransferases in C. glutamicum and it is unclear as to which ones are effective for increasing L-threonine production, (2) Katsumata et al. do not teach the type of aminotransferase used and do not teach the polypeptide of SEQ ID NO: 2, (3) Edwards et al. teach that aspartate aminotransferase is effective for increasing L-phenylalanine production in E. coli, (4) the L-threonine biosynthesis pathway is completely different from the L-phenylalanine pathway, and (5) Kishino et al. fail to make up for the deficiencies of Katsumata et al. Thus, it is Applicant's contention that the references, either alone or in combination, fail to teach the claimed invention.

11. Applicant's arguments have been fully considered but are not deemed persuasive to overcome the instant rejection or avoid the rejection of new claims 23-24. Claims 23-24 are directed to the methods of claims 12 and 21, respectively, with the added limitation that the modification that increases expression of the recited genes is the replacement of the endogenous promoter with a lac, trp, trc, PR, or PL promoter. With regard to arguments that Katsumata et al. do not teach the type of aminotransferase used, it is noted that Katsumata et al. teach that the aminotransferase used is an aspartate aminotransferase. See abstract

and page 1, lines 21-22 (called AAT throughout the entire reference). Thus, while the Examiner acknowledges Applicant's submission of a reference indicating that there are many types of aminotransferases, in the instant case, Katsumata et al. teach (1) a protein having the same enzymatic activity as that of the polypeptide of SEQ ID NO: 2, and (2) the increase in L-threonine production as a result of increasing the synthesis of an enzyme from *C. glutamicum* having aspartate aminotransferase activity. The *E. coli* strain of Debabov et al. is a high L-threonine producer which has been modified to increase the expression of a mutant *E. coli* thrA gene, the *E. coli* thrB gene, the *E. coli* thrC gene and the *E. coli* rhtA gene. In addition, as previously stated, aspartate aminotransferase catalyzes the conversion of oxaloacetate to aspartate, which is a precursor of L-threonine. Thus, in view of the fact that (1) the *E. coli* strain of Debabov et al. is able to produce L-threonine, (2) Katsumata et al. clearly teach that one could enhance the production of L-threonine by increasing the synthesis of aspartate aminotransferase, and (3) aspartate aminotransferase catalyzes the formation of a precursor of L-threonine, there is clear motivation to further modify the *E. coli* strain of Debabov et al. such that there is an increase in the production of aspartate aminotransferase for the benefit of further increasing L-threonine yields. One of skill in the art would be motivated to use the *E. coli* gene encoding the aspartate aminotransferase as this would not be seen as foreign to the *E. coli* strain of Debabov et al. Since the gene encoding the *E. coli* aspartate aminotransferase was known at the time the invention was conceived (taught by Edwards et al.) and the art (Edwards et al.) also taught overexpression of this gene for the production of other amino acids, one of skill in the art would have been clearly motivated to modify the *E. coli* strain of Debabov et al. such that the *E. coli* gene of Edwards et al. is overexpressed to increase the production of L-threonine. One of skill in the art would have a reasonable expectation of success at producing L-threonine from cultivation of the *E. coli* strain of Debabov further modified to increase the copy number of the gene of Edwards et al., or increase the expression of the gene of Edwards et al. by using a strong heterologous promoter in view of the fact that (1) the *E. coli* strain of Debabov et al. already produces high amounts of

L-threonine and there is no reason to believe that increasing the synthesis of the *E. coli* aspartate aminotransferase taught by Edwards et al. would impair the production of L-threonine already observed in the *E. coli* strain of Debabov et al., (2) Katsumata et al. shows that higher yields of L-threonine are observed when a gene encoding aspartate aminotransferase is overexpressed, and (3) techniques to increase the copy number of the *E. coli* gene of Edwards et al. are known in the art.

The use of low copy number vectors or the recited inducible promoters in claims 23-24 is also an obvious variation of the method of Debabov, Katsumata and Edwards. The teachings of Kishino et al. are further evidence that the use of low copy number vectors is obvious with regard to the production of L-threonine in a transformed *E. coli* cell. One of skill in the art would be motivated to use a low copy number vector or an inducible promoter in view of the fact that these would allow for better control of how much aspartate aminotransferase is produced and avoid intracellular instability. One of skill in the art would have a reasonable expectation of success at using low copy number vectors or use the specified inducible promoters in view of the fact that low copy number vectors and inducible promoters are well known and widely used in the art. Therefore, contrary to Applicant's assertions, the invention as a whole would have been prima facie obvious to a person of ordinary skill in the art at the time the invention was made.

***Conclusion***

12. No claim is in condition for allowance.

13. Applicant's amendment which added claims 23-24 necessitated the new ground(s) of rejection presented in this Office action. Accordingly, **THIS ACTION IS MADE FINAL**. See MPEP § 706.07(a). Applicant is reminded of the extension of time policy as set forth in 37 CFR 1.136(a).

A shortened statutory period for reply to this final action is set to expire THREE MONTHS from the mailing date of this action. In the event a first reply is filed within TWO MONTHS of the mailing

date of this final action and the advisory action is not mailed until after the end of the THREE-MONTH shortened statutory period, then the shortened statutory period will expire on the date the advisory action is mailed, and any extension fee pursuant to 37 CFR 1.136(a) will be calculated from the mailing date of the advisory action. In no event, however, will the statutory period for reply expire later than SIX MONTHS from the date of this final action.

14. Information regarding the status of an application may be obtained from the Patent Application Information Retrieval (PMR) system. Status information for published applications may be obtained from either Private PAIR or Public PAIR. Status information for unpublished applications is available through Private PAIR only. For more information about the PAIR system, see http://pair-direct.uspto.gov. Should you have questions on access to the Private PAIR system, contact the Electronic Business Center (EBC) at 866-217-9197 (toll-free).

15. Any inquiry concerning this communication or earlier communications from the examiner should be directed to Delia M. Ramirez whose telephone number is (571) 272-0938. The examiner can normally be reached on Monday-Friday from 8:30 AM to 5:00 PM. If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, Dr. Ponnathapura Achutamurthy can be reached on (571) 272-0928. Any inquiry of a general nature or relating to the status of this application or proceeding should be directed to the receptionist whose telephone number is (571) 272-1600.

Delia M. Ramirez, Ph.D.
Patent Examiner
Art Unit 1652

DR
January 4, 2007